

Relevance of bacterial secretion systems Type III and Type VI in the *Bradyrhizobium-Lupinus* symbiosis

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State of art and aim: One of the most studied models in plant-microbe interaction is the symbiosis *Rhizobium*-legume. This symbiosis is highly specific and depends on several molecular signals produced by both partners. Some of these signals are bacterial proteins named effectors that are translocated into the plant cells by secretion systems similar to contractile nanomachines (injectisomes). The injectisomes puncture and deliver the effectors into the target cell. The two main injectisomes are the secretion system type III (T3SS) and the secretion system type VI (T6SS). The genome of many rhizobia encodes T3SS and/or T6SS but their role in symbiosis is mostly unknown. The aim of this work is to study the symbiotic relevance of T3SS and T6SS of *Bradyrhizobium* that nodulate lupins that thrive in alkaline (*Lupinus mariae-josephae*) and acid soils (*L. angustifolius*) in the Iberian Peninsula.

Results and discussion: The analysis of the genomes of *Bradyrhizobium* strains: LmjC isolated from *L. mariae-josephae* and ISLU10 isolated from *L. angustifolius*, allowed the identification of one T3SS in the first strain and two T6SS in the second one. The genes of the secretion systems were grouped and those encoding structural components showed a high degree of conservation regarding the genes from other rhizobia. The strain LmjC is an efficient symbiont with *L. mariae-josephae* while it induces few ineffective and white nodules with *Lupinus angustifolius*. The T3SS of LmjC is encoded in a group of 30 genes. A mutant in the transcriptional regulator *ttsI*, essential for the activation of the T3SS, induces the formation of white nodules unable to fix nitrogen with *L. mariae-josephae* while *L. angustifolius* inoculation with the same mutant produces abundant red and effective nodules. These results suggest a key role of the T3SS of LmjC in defining the lupin host range of this bacterium.

The strain ISLU101 contains two clusters of genes involved in the formation of T6SS. One, T6SS-1, containing 17 genes showed a high degree of conservation regarding the genes of *B. japonicum* USDA110, a well known endosymbiont of soybean. The other, T6SS-2, contains 16 genes flanked by transposition sequences. This second gene cluster does not show high similarity to other rhizobia. Currently we are studying the role of these two T6SS in the symbiosis with *L. angustifolius* by generation of specific mutants.

References:

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